Observations on the *Haemoproteus* of Pigeons in Honolulu, Hawaii

L. KARTMAN[1]

DURING THE PERIOD January to July, 1948, observations were made at different times on the halteridium parasite of the pigeon, *Haemoproteus columbae* Kruse, 1890. Inasmuch as very little information has been published on the Haemosporidia of birds in Hawaii, these notes are presented with a view toward opening anew the question of blood protozoa of birds in the Territory, a problem which has never been systematically investigated. Fisher (1948) has emphasized the paucity of evidence regarding these parasites in Hawaii.

Alicata (1939, 1947) indicated that pigeons in Hawaii are commonly infected with *Haemoproteus columbae,* and Bryan (1934) showed that the hippoboscid vector, *Pseudolynchia canariensis* (Macq.), is generally distributed. No quantitative data are given in the above reports, however. The notes presented in this paper offer preliminary data on various aspects of the problem.

Pigeons and doves were examined at the Honolulu Zoo. Observations on both juvenile and adult pigeons showed them to be generally infested with the fly vector. There was an average of 2.0 flies per bird on about 50 juvenile pigeons examined and an average of 1.3 flies per bird on 100 adult pigeons. Of 45 doves examined none was found to harbor the pigeon fly, although the birds were housed in the vicinity of the pigeon lofts. This observation is in conformity with the remarks of Bequaert (1939), who indicated that *Pseudolynchia canariensis* has never been found on a wild host in North America. On the other hand, it has been taken from at least eight species of wild Columbidae belonging to five genera in Europe, Africa, and the Philippines.

More consistent infestation was noted on juvenile pigeons which had attained full plumage. Young birds between 15 and 25 days of age had the major infestation with flies whereas younger birds were less consistently infested. Many fly puparia were found in the pigeon nests and some of these were taken to the laboratory, placed in test tubes plugged with cotton, and kept at room temperature. Flies emerged from these pupae at from 15 to 20 days after collection. Pupae observed from the time of deposition by the female fly hatched at between 23 and 37 days.

Some of the pigeon flies collected at various times were dissected for evidence of infection with the pigeon *Haemoproteus*. Table 1 summarizes the data for dissections of 25 flies taken from both young and adult pigeons. A total of 36 per cent of the flies was found to be infected; it is interesting to note that of 19 females, 36.8 per cent were infected, and of 6 males, 33.3 per cent. Although flies from several juvenile pigeons were not infected, others collected from young birds showed the infection, indicating that the fly is active in migrating from adults to young.

Blood smears were taken from birds at the zoo to determine the incidence of infection with *Haemoproteus columbae* in pigeons and to learn whether local doves are naturally infected with the pigeon parasite. Although

[1] Department of Parasitology, University of Hawaii Agricultural Experiment Station. Manuscript received July 30, 1948.

TABLE 1

INFECTION OF PSEUDOLYNCHIA CANARIENSIS WITH HAEMOPROTEUS COLUMBAE IN NATURE

DATE	FLIES DISSECTED	FLIES INFECTED	NUMBER OF INFECTIONS		FLIES INFECTED	AGE OF HOST
			Midgut *	Salivary glands		
	Number	*Number*			*Per cent*	
3–30–48	12	5	5	2	41.7	juveniles and adults
3–31–48	4	0	0	0	0.0	juveniles only
5–12–48	9	4	4	2	44.4	juveniles and adults
Totals	25	9	9	4	36.0	

* In addition to typical oöcysts found on the midgut, it may be of interest to note oöcysts found on the hindgut of one fly (see Fig. 3).

blood smears cannot indicate the maximum rate of infection with *Haemoproteus,* they are a rapid and accurate method of surveying for approximate incidence of infection. The smears taken from pigeons and doves were subjected to Giemsa's stain, usually within 24 hours after they were taken, and were then examined. Besides recording the positive and negative smears, in every positive smear estimates were made of the number of gametocytes infecting red blood cells. In making this estimation the actual rate of infection in 2,000 or more red cells was first tabulated and then calculated in terms of gametocytes per 10,000 red cells. The data for these observations are shown in Table 2 and Figure 1. In addition to the data shown in the table, 38 juvenile pigeons under 30 days of age were all found negative. Two Chinese doves, *Streptopelia chinensis,* were also negative.

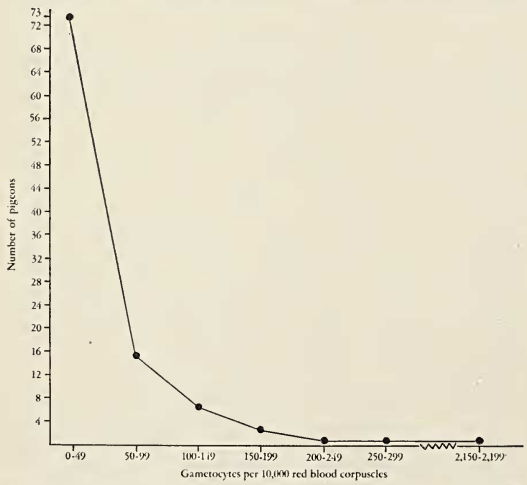

FIG. 1. Intensity of infection with *Haemoproteus columbae* in 101 pigeons (based on blood smears).

It is seen that the pigeons have a high rate of infection and that this rate is a little over two times the rate of infection found in the fly vector. Such an incidence in both definitive and intermediate hosts indicates an ideal situation for the life cycle of the parasite. It should be noted that most of the birds examined had mild infections. As was expected, none of the doves examined showed an infection with the pigeon parasite.

Several experiments were conducted in an attempt to transmit the *Haemoproteus* of pigeons to doves. It has been shown by Huff (1932) that the *Haemoproteus* of doves could be transmitted to domestic pigeons by the pigeon fly. However, the pigeon parasite has never been successfully transmitted to doves.

Experiment I: (1). On February 20, pigeon flies were collected from juvenile pigeons and 17 were placed in a cage with an infected pigeon showing 230 gametocytes per 10,000 red cells.

(2). On February 29, 10 flies were recovered from the pigeon and five were placed on a Chinese dove, *Streptopelia chinensis* (Scopoli). This dove had been captured on February 2, and blood smears taken on February 2, 15, 22, and 29 were all negative for

TABLE 2

INFECTION OF PIGEONS AND DOVES WITH HAEMOPROTEUS COLUMBAE IN NATURE

DATE	BIRD	NUMBER EXAMINED	NUMBER POSITIVE	PERCENTAGE POSITIVE
1–20–48	Pigeon	3	3	100.0
2–23–48	Pigeon	28	20	71.4
2–25–48	Pigeon	45	42	93.3
2–26–48	Pigeon	25	18	72.0
3–30–48	Dove*	43	0	0.0
Totals (Pigeons only)		101	83	82.2

* Thirty Indian ring-neck doves and 13 lace-neck doves.

blood parasites. The remaining five flies not placed on the dove were dissected and four were found to have oöcysts on the midgut. Salivary gland examinations were not made.

(3). Blood smears taken on March 12 and 16 were negative and on March 29 the dove was found dead in its cage. Blood smears from the heart and liver were negative for *Haemoproteus* gametocytes.

(4). Five flies were recovered from the cage containing the dead dove and dissections of these showed two with both oöcysts and sporozoites, one with oöcysts only, and two with negative midguts and salivary glands.

Experiment II: (1). On May 12, pigeon flies were collected from juvenile pigeons. Of three flies dissected, one was positive for sporozoites. Fifteen flies were comminuted with physiological saline in a mortar, yielding 1.5 ml. of pooled material. Approximately 0.5 ml. was injected into the pectoral muscles of one ring-neck dove, *Streptopelia decaocto* (Privalszky), one barred dove, *Geopelia striata striata* (Linn.), and one 5-week-old white Leghorn chick.

(2). Blood smears taken from these birds on June 8, 17, and July 2 were all negative and the experiment was terminated.

Experiment III: (1). On May 12, 10 pigeon flies were placed on an infected pigeon showing 250 gametocytes per 10,000 red cells.

(2). On May 22 the flies were removed from the pigeon and placed on a ring-neck dove. Two flies dissected were found positive for oöcysts.

(3). Blood smears taken on June 17 and July 4 were negative. Six flies taken from the dove on July 4 showed four positive for sporozoites.

All the doves were examined well within the period shown by Adie (1924) to be necessary for the appearance of gametocytes in the peripheral blood. Even in the case of the dove in Experiment I a period of 30 days intervened between first feeding of the flies and death of the bird. Since infected flies were known to be present in these samples and since the flies feed on their host every day, it is felt that the doves had ample opportunity to become infected.

The failure to infect these doves and the chicken with the *Haemoproteus* from the pigeon substantiates the findings of Coatney (1933), who showed that the mourning dove, *Zenaidura macroura carolinensis,* is not susceptible to *Haemoproteus columbae* of the pigeon. He ascribes this to the known high degree of host specificity among many of the Haemosporidia and concludes that doves have a high degree of natural immunity toward the pigeon *Haemoproteus.*

Although earlier in this paper it was indicated that the pigeon fly was not taken on doves in nature, no difficulty was encountered in using the doves as hosts of this vector in the laboratory. Coatney (1933) also found this to be true in the case of the mourning dove.

Various workers have indicated that early stages in the sporogony of *Haemoproteus columbae* are capable of a certain amount of development in bloodsucking insects other than the hippoboscid vector. Wenyon (1926: 897) has summarized the reports of Aragão and Nöller, who indicated that oökinetes were formed in mites, bedbugs, and various species of culicine mosquitoes. In *Aëdes argenteus* (=*Aëdes aegypti*) Nöller found that oökinetes

formed quite readily at a temperature of 11° to 12° C. and persisted for at least 6 days.

Several experiments were conducted to test the developmental ability of the pigeon *Haemoproteus* in local mosquitoes. In all cases the mosquitoes were reared from larvae and pupae collected in the field. Adult females were fed on moist raisins for at least 2 days and were then kept from both food and water for another 2 days before their first blood meal. They were liberated into a cage 30 inches high, 28 inches wide, and 32 inches deep for feeding on infected pigeons. Feathers were plucked from about the body of the pigeons to expose the skin and to allow greater opportunity for engorgement by the mosquitoes. No difficulty was encountered in getting a sufficient number of engorged mosquitoes for the observations described below, although apparently many of the females did not feed on the pigeons. All experiments were conducted at room temperature, which fluctuated between 62.5° and 83.5° F. during this period.

Experiment I: (1). On February 5, starved and thirsty females of *Culex quinquefasciatus* Say were liberated into the cage containing an infected pigeon with 250 gametocytes of *Haemoproteus columbae* per 10,000 red cells. The mosquitoes were allowed to feed overnight and engorged females were collected the next morning, placed in a small cage, and maintained on moist raisins. The engorged individuals were presumed to have had one blood meal during the night.

(2). Ten females were dissected on February 15, 10 on February 17, 5 on February 21, and 10 on March 10. All of these examinations showed midguts negative for *Haemoproteus* oöcysts.

Experiment II: (1). On April 30, starved and thirsty females of *C. quinquefasciatus* were liberated into a cage with a pigeon showing 175 gametocytes per 10,000 red cells. These mosquitoes were allowed to remain in the cage with the pigeon throughout the experiment and some of the engorged individuals presumably had more than one blood meal.

(2). Fifteen females were dissected on May 9, and 18 on May 16 with completely negative findings.

Experiment III: (1). On May 8, 10 starved and thirsty *C. quinquefasciatus* females were liberated into a cage with a pigeon showing 250 gametocytes per 10,000 red cells.

(2). On May 9, after about 18 hours, all of these females were dissected with the following results: four females showed no evidence of having taken a blood meal; six females were partially to completely engorged and stomach smears treated with Giemsa's stain showed many gametocytes of *Haemoproteus columbae* still within red cells and some exoerythrocytic rounded macrogametocytes. Some extracellular

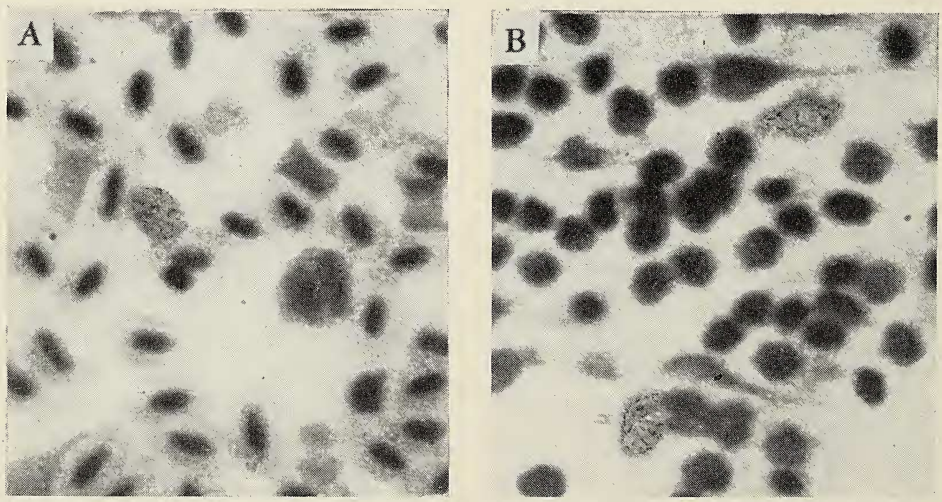

FIG. 2. Globular forms of macrogametocytes of *Haemoproteus columbae* after breaking out of red cells. (A) Smear of heparinized blood from infected pigeon. Note that one gametocyte is just breaking away from red cell. (B) Smear from midgut of *Aedes albopictus* one hour after engorging on infected pigeon. (Smears treated with Giemsa's stain. Photomicrograph by Dr. G. B. Mainland; slightly retouched.)

microgametocytes were noted, but a diligent search failed to show definite evidence of exflagellation.

All the rounded gametocytes were morphologically similar to those found in smears made from heparinized pigeon blood (see Fig. 2). No oökinetes were seen and most of the gametocytes were intracellular and typical halter forms, indicating probable immaturity.

Experiment III also included observations on *Aëdes albopictus* (Skuse). Starved and thirsty females of this species were liberated into the cage on May 8 along with the other culicine described above. Two of these females were dissected 1 hour after feeding on the infected pigeon and stomach smears showed a few rounded macrogametocytes. On May 9, about 20 hours after being placed in the cage, five engorged females were dissected. Stomach smears again revealed some rounded macrogametocytes. Ten additional females were dissected on May 15 and 15 more were examined on May 23. All midguts were negative for oöcysts.

As in the case of *Culex quinquefasciatus,* the stomach smears of *A. albopictus* showed a few rounded macrogametocytes, some extracellular microgametocytes, and many halter forms still within intact red cells. No oökinetes were noted.

Experiment IV: An attempt was made to infect the hippoboscid fly (*Olfersia aenescens* Thomson), which is normally found on ocean birds. The flies used were collected by employing a juvenile red-footed booby, *Sula sula rubripes,* as a decoy. About 36 *O. aenescens* were brought to the laboratory on February 28. Ten were dissected the next day and were found negative for evidence of oöcysts on the midgut. On February 29, 20 of these flies were liberated in a cage with an infected pigeon showing 230 gametocytes of *Haemoproteus columbae* per 10,000 red cells. Most of the flies seemed attracted to the lighter side of the cage and were not apparently interested in the pigeon. Several flies were found dead on each succeeding day and none was seen flying about the cage after 7 days. On March 10 the pigeon was removed from the cage and only one *O. aenescens* was recovered from it. Dissection of this fly showed fresh blood in the midgut, but there was no evidence of oöcyst formation.

Experiment V: Four puparia were collected from the captive *Olfersia aenescens* described above. At room temperature two of these hatched in from 52 to 53 days, but the others failed to emerge. The two flies were placed on an infected pigeon on April 24, and both immediately ran underneath the feathers. On May 4 these two flies were taken from the bird and dissected with negative findings.

SUMMARY

1. Observations have been made on the halteridium parasite of the pigeon, *Haemoproteus columbae* Kruse, 1890, in pigeons from lofts at the Honolulu Zoo.
2. The hippoboscid vector, *Pseudolynchia canariensis* (Macq.), was found to be present at an average rate of 2.0 per bird on about 50 juvenile pigeons and 1.3 per bird on 100 adult pigeons. Of 45 doves examined, none harbored this fly.
3. Of a total of 25 *P. canariensis* dissected, 9 or 36.0 per cent were found to be naturally infected with the pigeon *Haemoproteus.*
4. Of 101 adult pigeons examined by the blood smear technique, 83 or 82.2 per cent were positive for *Haemoproteus columbae.* Of 43 doves examined, none was found infected with the pigeon parasite or any other blood protozoan.

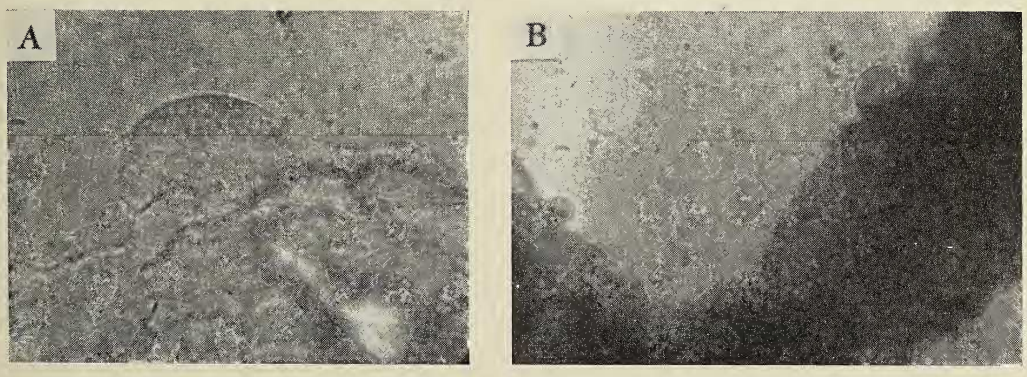

FIG. 3. Left: Oöcyst of *Haemoproteus columbae* on midgut of *Pseudolynchia canariensis.* Approx. 950×. Right: Oöcysts of *H. columbae* on hindgut of *P. canariensis.* Approx. 440×.

5. Attempts to transmit the pigeon *Haemoproteus* to three species of doves and to a young chicken, either by means of the fly vector or by inoculation of macerated flies, proved negative.
6. Oökinetes and oöcysts failed to develop in *Culex quinquefasciatus* Say, *Aëdes albopictus* (Skuse), and *Olfersia aenescens* Thomson when these species were allowed to feed on infected pigeons. *O. aenescens* did not feed readily on pigeons. Smears of the stomach contents of engorged female mosquitoes of both species showed mainly halter forms in red cells and some rounded macrogametocytes. Some extracellular microgametocytes were also noted but definite evidence of exflagellation was not found.

Acknowledgments: Thanks are due the following persons for their aid in these experiments: Paul Breese, Director of the Honolulu Zoo, for generously allowing complete freedom in observations and examinations of pigeons and other birds; Jane Y. C. Yuen for her aid in taking blood smears and for laboratory help; Dr. D. D. Bonnet, Territorial Board of Health, for supplies of larval and pupal *Culex quinquefasciatus;* Dr. Frank Richardson, Assistant Professor of Zoology, University of Hawaii, for help in collecting hippoboscid flies from ocean birds; and Dr. J. Bequaert, Harvard Museum of Comparative Zoology, for identifying the *Olfersia aenescens.* Appreciation is also due the University of Hawaii Agricultural Experiment Station for the use of certain of its facilities.

REFERENCES

ADIE, H. A. 1924. The sporogony of *Haemoproteus columbae. Soc. Path. Exot., Bul.* 17(7): 605–613.

ALICATA, J. E. 1939. Poultry parasites. *Hawaii Agr. Expt. Sta. Rpt. 1938:* 79–82.

——— 1947. Parasites and parasitic diseases of domestic animals in the Hawaiian Islands. *Pacific Sci.* 1(2): 69–84.

BEQUAERT, J. 1939. Hippoboscid flies from North American doves. *Science* 89 (2308): 267–268.

BRYAN, E. H., JR. 1934. A review of the Hawaiian Diptera, with descriptions of new species. *Hawaii. Ent. Soc., Proc.* 8(3): 399–468.

COATNEY, G. R. 1933. Relapse and associated phenomena in the *Haemoproteus* infection of the pigeon. *Amer. Jour. Hyg.* 18(1): 133–160.

FISHER, H. I. 1948. The question of avian introductions in Hawaii. *Pacific Sci.* 2(1): 59–64.

HUFF, C. G. 1932. Studies on *Haemoproteus* of mourning doves. *Amer. Jour. Hyg.* 16(2): 618–623.

WENYON, C. M. 1926. *Protozoology.* Vol. II. ix + 784 pp. 128 figs. Bailliere, Tindall and Cox, London.